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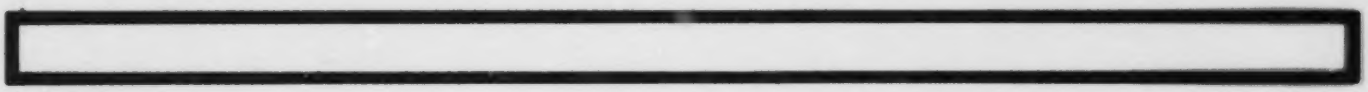

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16. ABSTRACT

A committee of scientists was established by the U.S. Environmental Protection Agency to appraise the current state of aneuploidy test methodology, to compile and analyze published data on the chemical induction of aneuploidy, and to provide guidance for additional test development and validation. The committee was organized into 9 working groups, representing different eukaryotic organisms ranging from fungi to whole mammals. The Environmental Mutagen, Carcinogen, and Teratogen Information Program at Oak Ridge National Laboratory identified and provided literature to the working groups. The 9 reports were prepared by the individual working groups. The reports include: (1) descriptions of the available testing approaches and general principles to be followed for protocol development, (2) criteria for evaluating data, and (3) recommendations for future test development and basic research. The evaluated results are presented in tables. For the positive and negative calls, the lowest effective dose tested or the highest ineffective dose tested, respectively, and the range of concentrations tested are provided. An overall summary report was prepared that summarizes the conclusions and recommendations of the 9 working groups and the chemical data base. It is hoped that these reports will provide a framework for further test development and validation of aneuploidy assays.

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Aneuploidy Data Review Committee: Summary compilation of chemical data base and evaluation of test methodology *

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The Aneuploidy Data Review Committee is composed of 9 working groups on different aneuploidy test systems (Dellarco et al., 1986); the reports of these working groups comprise the major part of this special issue of *Mutation Research*. This paper summarizes the conclusions of the committee and the data base of chemical test results. The data base on aneuploidy induction is too small at present for detailed comparisons of assay systems or assessments of chemical activity, but this systematic, critical reveiw of methodology provides guidelines for further test development and validation.

(1) Test methods

The occurrence of aneuploidy can be measured in diverse eukaryotic organisms: fungi, plants, insects, and mammals. In fungi and Drosophila, the end point detected in aneuploids is the loss or expression of specially marked chromosomes (Resnick et al., 1986; Käfer et al., 1986; Griffiths et al., 1986; Zimmering et al., 1986). Because in these systems clastogenic or recombinogenic agents can produce phenotypes similar to those expected for aneuploids, special test strains have been developed to distinguish aneuploidy from other possible effects. Test protocols have been described by the working groups for Neurospora (Griffiths et al., 1986) and Aspergillus (Käfer et al., 1986). In Aspergillus, it is possible to distinguish between aneuploidy induced as a primary effect on the mitotic apparatus and those derived secondarily from structural chromosome aberrations. Improved strains for aneuploidy detection in Saccharomyces and Drosophila have been constructed recently (Resnick et al., 1986; Zimmering et al., 1986), and protocols using several of these strains are under validation.

Plant systems for aneuploidy detection usually have cytogenetic end points; meiotic (microsporocyte) and mitotic (root tip) tissues can be used. A mitotic marker segregation assay is available in soybeans (Vig, 1975), and a system in wheat is being developed (Redei et al., 1985). To date however, most of the work on plant aneuploidy has been qualitative, for purpose of genetic mapping or improvement of agricultural varieties. Protocols for quantifying aneuploidy induction in meiotic

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and mitotic cells are described by the working group, but experience in the use of the assays for this purpose is needed.

Most of the published data on aneuploidy induction in mammalian somatic cells consists of ancillary observations from studies of structural chromosomal aberrations. Reports of the working groups on cultured cells (Galloway and Ivett, 1986) and somatic cells in vivo (Cimino et al., 1986) outline basic steps to be included and some pitfalls to be avoided in developing protocols specifically for aneuploidy detection. Work on new assay systems, both in vitro and in vivo, is underway in several laboratories (Athwal and Sandhu, 1985; Ivett and Brown, 1985; Liang and Satya-Prakash, 1985; Tanzarella et al., 1985; Tsutsui et al., 1984).

Chromosome counts in metaphase II spermatocytes or in embryos fathered by treated males are presently the most reliable cytogenetic methods for detecting aneuploidy in male germ cells (Allen et al., 1986). Many papers report analysis of 'related' end points in germ cell stages other than metaphase II (e.g., univalents in metaphase I or fluorescent sex-chromatin bodies in postmeiotic cells); the postulated connection between these end points and actual aneuploid chromosome counts in germ cells or progeny has been questioned with increasing vigor for several years (Liang and Hsu, 1985; Wyrobek et al., 1984; Sugawara and Mikamo, 1983). The working group on male germ cells has recommended preliminary guidelines for development of cytogenetic assays and has discussed some promising new methods (Allen et al., 1986).

Chromosomal complements of human spermatozoa can now be studied after in vitro fertilization of hamster oocytes (Martin, 1984; Brandriff et al., 1984). This approach permits accurate determination of the background rates of aneuploidy in human spermatogenesis and a method for assessing risk to exposed human populations. Similar assays using sperm of treated animals could be developed for the screening of chemicals.

Chromosome counts in metaphase II oocytes of mammals have provided more reliable data than the analogous test in spermatocytes. Adequate sample sizes are obtained more conveniently if the animals are superovulated, but the effects of this treatment on the background frequency of aneuploidy should be investigated more thoroughly. Chromosome counts in pronuclei at first cleavage can be used to measure aneuploidy induced in germ cells of either sex, but most studies of this type have used treated females. The working group report on female germ cells (Mailhes et al., 1986) gives detailed protocols for assays using metaphase II oocytes or fertilized eggs of mice; adaptations will be necessary if other species are used. Comparative studies of different mammalian species and laboratory strains would be helpful.

Genetic as well as cytogenetic methods have also been proposed to detect aneuploidy arising in the germ cells of mice, using sex chromosome loss (Russell, 1976, 1979) or tester stocks bearing Robertsonian translocations (Cattanach et al., 1984); no chemical test data have been published. A new technique, which scores trisomic offspring of treated mice as late fetal deaths, was recently described (Russell, 1985).

None of the aneuploidy assay systems have been sufficiently tested and validated. Neurospora, Aspergillus, and some Drosophila systems are ready for validation; those using Saccharomyces, newer Drosophila strains, some plants (Allium, Vicia, Hordeum), or mouse oocytes could be readied for validation in a relatively short time. For most assays using mammalian material, only the outlines of possible protocols are discernible at this time. Tests using intact animals may be too costly and time consuming for the primary screening of chemicals, but it is against data from mammalian tests that the faster, less expensive assays must be validated. The development of mammalian assays and the generation of a reliable data base of test results for mammals are goals with high priority.

(2) General guidelines for protocol development

A large fraction of the experimental data reviewed by the working groups was judged to be inconclusive. Government agencies and the private sector rely heavily on published data to determine the toxicological potential of chemicals, and it is clear that many of the published data on the chemical induction of aneuploidy are inadequate. Some deficiencies in the literature apply across different test systems; lack of control data, inade-

quate sample sizes, improper protocols, or insufficient reporting of procedures or test results. These inadequacies severely limit the usefulness of the current data base, but they do provide guidance for future experimental design.

(A) The end point measured is to be a reliable indicator of aneuploidy, as shown by actual chromosome counts or unequivocal genetic evidence for the loss or gain of whole chromosomes. Other end points (e.g., lagging chromosomes, premature centromere division, or binding of chromosome-specific-labeled DNA probes) may be predictive of aneuploidy and useful indicators of aneuploidy induction, but the relationship must be carefully documented, not assumed.

(B) Negative and positive controls are essential, but reliable positive control compounds have not been identified for most of the aneuploidy assays. Because there may be several different intracellular targets for aneuploidy-inducing agents (see Mason and Resnick, 1985; Liang and Brinkley, 1985; Dellarco et al., 1985; Bond and Chandley, 1983), several different positive control compounds may be necessary. Some solvents have been shown to induce aneuploidy, including ethanol (Käfer, 1984; Zimmermann et al., 1985), acetone (Zimmermann et al., 1984, 1985), and dimethyl sulfoxide (Fulton and Bond, 1984; Zimmering and Mason, personal communication; Zimmermann and Resnick, personal communication). Solvent-treated controls are thus a necessity, and solvent effects on aneuploidy induction by test agents should be considered.

(C) Sample size should be adequate to support the chosen method of statistical analysis, given the background frequency of the assay system and the desired level of significance.

(D) Sampling times should be chosen with regard for the replication kinetics of the cells used. Completion of at least one cell cycle after treatment is necessary for production of aneuploids, but prolonged growth increases the chance for death of or selection against aneuploid products.

(E) When assays are carried out in culture media, the physical conditions need careful attention. Temperature, pH, and perhaps other factors, affect the background frequency of aneuploids (Galloway and Ivett, 1986). Sometimes these factors can be exploited; an incubation period in the cold has been used to increase the sensitivity of the Saccharomyces assay (Zimmermann et al., 1985). In all cases, conditions should be controlled and consistent for all treatment groups.

(F) Several different dose levels should be tested. It seems likely that some aneuploidy-inducing chemicals will be active over a narrow concentration range (Brinkley et al., 1985). For a negative test to be valid, the highest dose tested is to produce some evidence of toxicity.

(G) Toxicity data should be reported along with aneuploidy data.

(H) Test procedures and results should be reported in sufficient detail for independent analysis and evaluation by the reader. Specific recommendations for data presentation are given in the working group reports. For cytogenetic tests, it is important to report hyperploidy and hypoploidy as separate data categories.

(3) Test results

Table 1 lists the chemicals evaluated by the 9 working groups and the positive, negative or inconclusive assessments for each chemical (with positives and negatives, the number of corroborating reports from different studies is given). For more than one-half of the chemicals listed, all results were judged inconclusive (reasons for inconclusive assessments are given in the working group reports). In an overwhelming majority of the cases where results could be accepted as positive or negative, only one independent report was acceptable for a particular compound in a particular assay system. These data were judged to be reliable but need confirmation. Moreover, of the compounds evaluated as negative or positive, 75% have positive or negative results in one assay system *only*. Table 1 presents evaluations of particular papers or studies. Interpretation of the information in Table 1 as definitive assessments of a particular chemical's activity would exceed both the capacity of the data to support the interpretation and the intention of the Aneuploidy Data Review Committee in presenting it.

Various environmental and therapeutic agents have given positive results in at least one aneuploidy test system (Table 1). Hormones, pesticides, antibiotics, antineoplastic drugs and solvents are

TABLE 1

SUMMARY OF TEST RESULTS ON CHEMICALS COVERED BY THE ANEUPLOIDY DATA REVIEW COMMITTEE [a]

Chemical (CAS Registry No.)	Test system [b]								
	Sacch.	Asper.	Neuro.	Plant	Droso.	Mam-CC	Mam-SC	Mam-FG	Mam-MG
Acenaphthene (83-32-9)					+(1) [c]				
Acephate (30560-19-1)			−(1)						
Acetin (26446-35-5)					I				
Acetone (67-64-1)	+(1)								
2-Acetylaminofluorene (53-96-3)	I		−(1)				I		
4-Acetylaminofluorene (28322-02-3)	I								
Acridine orange (65-61-2)			−(1)						
Acriflavine (8048-52-0)								I	
Acrylamide (79-06-1)							I		I
Actinomycin D (50-76-0)		+(1)			+(1)				
Adenosine triphosphate (56-65-5)					I				
Adriamycin (23214-92-8)	I		I	I					I
Aflatoxin B_1 (1162-65-8)							I		
Alloxan (50-71-5)			−(1)				I		
Allyl glycidyl ether (106-92-3)					I				
α-Amanitin (23109-05-9)					I				
Ametryne (834-12-8)			−(1)						
2-Aminofluorene (153-78-6)			−(1)						
2-Amino-4-nitrophenol (99-57-0)			I						
2-Amino-5-nitrophenol (121-88-0)			I						
3-Aminophenol (591-27-5)			+(1)						
4-Aminophenol (123-30-8)			I						
Aminopterin (54-62-6)			−(1)						
Amitrole (61-82-5)	I	W(1)	−(1)		−(1)				
Amphotericin B (1397-89-3)	I	+(1)	−(1)						

TABLE 1 (continued)

Chemical (CAS Registry No.)	Test system [b]								
	Sacch.	Asper.	Neuro.	Plant	Droso.	Mam-CC	Mam-SC	Mam-FG	Mam-MG
Anthracene (120-12-7)	I								
Anthralin (1143-38-0)	I								
Arsenic (7440-38-2)					I		I		
Asbestos (chrysotile) (12001-29-5)						+(1)			
Ascorbic acid (50-81-7)	I								I
Aspirin (50-78-2)				+(1)					
Atenolol (29122-68-7)	I								
Atrazine (1912-24-9)			+(1)	+(2)	−(1)				
Auramine (2465-27-2)	I								
5-Azacytidine (320-67-2)	−(1)								
8-Azaquanine (134-58-7)			I						
Azathioprine (446-86-6)					+(1)		I		
Azoxybenzene (495-48-7)	I								
Azure A (531-53-3)			−(1)						
Azure B (531-55-5)			−(1)						
Azure C (531-57-7)			−(1)						
Benomyl (17804-35-2)	+(1)	+(4)	−(1)		−(1)	+(1)			I
Benz[*a*]anthracene (56-55-3)						I			
Benzene (71-43-2)							I		
Benzidine (92-87-5)	I								
Benzo[*a*]pyrene (50-32-8)	I		−(1)	−(1)		+(1)	I	I	
Benzyl chloride (100-44-7)	I								
Bleomycin (11056-06-7)	I	−(1)			+(1)		I		I
Botran (99-30-9)		+(1)	−(1)						
Brilliant blue (2650-18-2)			I						
Bromacil (314-40-9)			+(1)						
Busulfan (55-98-1)									I
Butylated hydroxyanisole (25013-16-5)					I				

TABLE 1 (continued)

Chemical (CAS Registry No.)	Test system [b]								
	Sacch.	Asper.	Neuro.	Plant	Droso.	Mam-CC	Mam-SC	Mam-FG	Mam-MG
Butylated hydroxytoluene (128-37-0)					I				
Butylnitrosourea (869-01-2)							I		
γ-Butyrolactone (96-48-0)	I								
Cadmium (7440-43-9)							I		
Cadmium chloride (10108-64-2)								+(3)	I
Cadmium sulfate (10124-36-4)							I		
Caffeine (58-08-2)	I		+(1)		+(2)				
Calcium chromate (13765-19-0)					i				
trans-Cannabidiol (13956-29-1)							I		
Cannabinol (521-35-7)							I		I
Captan (133-06-2)		−(1)					I		
Carbaryl (63-25-2)						+(1)			I
Carbon disulfide (75-15-0)							I		
Carboxin (5234-68-4)		W(1)							
Chenodeoxycholic acid (474-25-9)	I								
Chloral hydrate (302-17-0)		+(2)	I						+(1)
Chloramphenicol (56-75-7)				+(1)					
Chlorfurazol (3615-21-2)		−(1)							
Chloroform (67-66-3)	I								
4-Chloromethylbiphenyl (1667-11-4)	I							−(1)	I
Chloroneb (2657-77-6)		+(2)							
4-Chlororesorcinol (95-88-5)			I						
4-Chloro-2-tolyloxyacetic acid (94-74-6)				−(1)					
Chlorpropham (101-21-3)	I	+(1)							
Cholic acid (81-25-4)	I								
Clophen A30 (54991-93-4)					−(1)				

TABLE 1 (continued)

Chemical (CAS Registry No.)	Test system [b]								
	Sacch.	Asper.	Neuro.	Plant	Droso.	Mam-CC	Mam-SC	Mam-FG	Mam-MG
Clophen A50 (8068-44-8)					+(1)				
Cobalt chloride (7646-79-9)			I						
Colcemid (477-30-5)					+(1)	+(7)			I
Colchicine (64-86-8)			I	+(4)	I			+(1)	
m-Cresol (108-39-4)			I						
Croton oil (8001-28-3)			I						
Cyanazine (21725-46-2)			I		−(1)				
3′,5′-Cyclic AMP (60-92-4)			−(1)						
Cycloheximide (66-81-9)			I	W(1)					
Cyclohexylamine HCl (4998-76-9)							I		
Cyclophosphamide (50-18-0)	+(1)	+(1) [d]	−(1)		I			+(2)	+(1)
Cyproheptadine HCl (969-33-5)						−(1)			
Cytochalasin B (14930-96-2)			I						
p,*p*′-DDT (50-29-3)					+(1)		I		
Deoxycholic acid (83-44-3)	I								
2,7-Diacetylaminofluorene (304-28-9)							I		
2,4-Diaminoanisole sulfate (39156-41-7)			+(1)						
2,5-Diaminoanisole sulfate (66671-82-7)			I						
2,4-Diaminotoluene (95-80-7)			−(1)						
2,5-Diaminotoluene sulfate (6369-59-1)			I						
Diamminedichloro-dinitroplatinum (18726-50-6)				I					
cis-Diamminedichloro-platinum II (15663-27-1)					−(1)				
Diazepam (439-14-5)						+(1)			
Diazinon (333-41-5)			−(1)						

TABLE 1 (continued)

Chemical (CAS Registry No.)	Test system [b]								
	Sacch.	Asper.	Neuro.	Plant	Droso.	Mam-CC	Mam-SC	Mam-FG	Mam-MG
1,2-Dibromo-3-chloropropane (96-12-8)					I				I
2,3-Dibromopropanol (96-13-9)					I				
Dichloroacetonitrile (3018-12-0)					I				
1,2-Dichloroethane (107-06-2)					+(1)				
2,4-Dichlorophenol (120-83-2)				−(1)					
2,4-Dichlorophenoxy-acetic acid (94-75-7)				W(1)					
Dichlorvos (62-73-7)		+(1)							
1,2:3,4-Diepoxybutane (1464-53-5)		+(1)			I				
Diethyl pyrocarbonate (1609-47-8)				+(1)					
Diethyl sulfate (64-67-5)					I				
Diethylmercury (627-44-1)							I		
Diethylnitrosamine (55-18-5)					I		I		
Diethylstilbestrol (56-53-1)	I		−(1)			+(1)	I		
Diethylstilbestrol diphosphate (522-40-7)			−(1)				I		
Diethylstilbestrol dipropionate (130-80-3)	I								
15,16-Dihydro-11-methylcyclopenta[*a*]-phenanthren-17-one (5836-85-1)	I								
Dihydroxyurea (686-68-0)					I				
Dimethyl sulfate (77-78-1)					I				
4-Dimethylamino-azobenzene (60-11-7)	I						I		
4-Dimethylamino-2-methylazobenzene (54-88-6)							I		
4-Dimethylamino-3′-methylazobenzene (55-80-1)							I		
9,10-Dimethyl-anthracene (781-43-1)	I								

TABLE 1 (continued)

Chemical (CAS Registry No.)	Test system [b]								
	Sacch.	Asper.	Neuro.	Plant	Droso.	Mam-CC	Mam-SC	Mam-FG	Mam-MG
9,10-Dimethyl-1,2-benz-anthracene (57-97-6)						+(1)			
Dimethylcarbamyl chloride (79-44-7)	I								
Dimethylformamide (68-12-2)	I								
Dimethylnitrosamine (62-75-9)					I		I		
Dimethylsulfoxide (67-68-5)	I	−(2)	I		−(1)		I		
Dinitrosopenta-methylene tetramine (101-25-7)	I								
2,4-Dinitrotoluene (121-14-2)							I		
Diphenylmercury (587-85-9)							I		
Diphenylnitrosamine (86-30-6)	I								
Econazole (27220-47-9)		+(1)							
Epichlorohydrin (106-89-8)	I								
Eptam (759-94-4)			−(1)						
β-Estradiol (50-28-2)						I			
β-Estradiol dipropionate (113-38-2)							I		
Ethanol (64-17-5)		+(3)	−(1)				I	I	I
Ethidium bromide (1239-45-8)		−(1)							
DL-Ethionine (67-21-0)	I		−(1)						
Ethosuximide (77-67-8)						I			
Ethyl methanesulfonate (62-50-0)	+(1)		I	+(1)	I				I
Ethyl thiophanate (23564-06-9)		+(1)							
Ethylene glycol (107-21-1)			−(1)						
Ethylenethiourea (96-45-7)	I					−(1)	I		
Ethylnitrosourea (759-73-9)					I				
Fenarimol (60168-88-9)		+(1)							

TABLE 1 (continued)

Chemical (CAS Registry No.)	Test system [b]								
	Sacch.	Asper.	Neuro.	Plant	Droso.	Mam-CC	Mam-SC	Mam-FG	Mam-MG
Ferric chloride (7705-08-0)			I						
5-Fluorodeoxyuridine (50-91-9)		I	−(1)		+(1)				
p-Fluorophenylalanine (60-17-3)	+(1)	+(2)	+(1)						I
3-Fluoropyruvic acid (433-48-7)			I						
4-Fluorotryptophan (25631-17-8)			−(1)						
m-Fluoro-DL-tyrosine (403-90-7)			I						
5-Fluorouracil (51-21-8)			I						
Folic acid (59-30-3)			I						
Formaldehyde (50-00-0)			−(1)						I
N-Furfuryladenosine (4338-47-0)				I					
Fruosemide (54-31-9)									I
Fusarium T-2 toxin (21259-20-1)					−(1)				
Glutethimide (77-21-4)					−(1)				
Glycodeoxycholic acid (360-65-6)	I								
Griseofulvin (126-07-8)		+(1)		+(1)		+(1)			
Haloprogin (777-11-7)			−(1)						
Halothane (151-67-7)				I	+(1)			−(1)	I
Heliotrine (303-33-3)					−(1)				
Heroin (561-27-1)							I		
Hexamethylphosphoramide (680-31-9)	I								
Hycanthone (3105-97-3)			I						
Hycanthone methanesulfonate (23255-93-8)					I				
Hydrazine sulfate (10034-93-2)	I								
Hydroxylamine (7803-49-8					I				
4-Hydroxymethylbiphenyl (3597-91-9)	I								

TABLE 1 (continued)

Chemical (CAS Registry No.)	Test system [b]								
	Sacch.	Asper.	Neuro.	Plant	Droso.	Mam-CC	Mam-SC	Mam-FG	Mam-MG
8-Hydroxyquinoline (148-24-3)			I						
Ifosfamide (3778-73-2)					I				
Iodoacetic acid (64-69-7)	I								
Iodochlorhydroxyquin (130-26-7)		+(2)							
Isoniazid (54-85-3)								−(1)	−(1)
Isopropanol (67-63-0)			I						
Isopropyl methanesulfonate (926-06-7)					I				
Lasiocarpine (303-34-4)					I				
Lauric acid (143-07-7)	I								
Lead (7439-92-1)							I		
Lead nitrate (10099-74-8)							I		
α-Lindane (319-84-6)							I		
Lithocholic acid (434-50-5)	I								
Lucanthone (479-50-5)			I						
Lysergic acid diethylamide (50-37-3)					I		I		I
Malathion (121-75-5)			I						I
Maleic hydrazide (123-33-1)			−(1)				I		
Mecoprop (93-65-2)		−(1)							
Meprobamate (57-53-4)					I				
Merbromin (129-16-8)	I								
6-Mercaptopurine (50-44-2)							I		I
Mercuric chloride (7487-94-7)			I				I	−(1)	
Mercury (7439-97-6)							−(2)		
Mescaline (54-04-6)							I		
Methadone (76-99-3)									I

TABLE 1 (continued)

Chemical (CAS Registry No.)	Test system [b]								
	Sacch.	Asper.	Neuro.	Plant	Droso.	Mam-CC	Mam-SC	Mam-FG	Mam-MG
Methanol (67-56-1)			I						
l-Methionine (63-68-3)	I						I		
Methotrexate (59-05-2)			+(1)					+(2)	
Methoxychlor (72-43-5)									I
2-Methoxyethylmercury chloride (123-88-6)					−(1)				I
Methoxyflurane (76-38-0)				I					
8-Methoxypsoralen (298-81-7)			I						
Methyl 2-benzimidazolecarbamate (10605-21-7)	+(2)	+(1)					I		I
Methyl methanesulfonate (66-27-3)	W(1)	+(2)			I				I
Methyl orange (547-58-0)	I								
Methyl thiophanate (23564-05-8)		+(1)							
p-Methylaminophenol sulfate (55-55-0)			I						
4,4′-Methylenebis-(2-chloroaniline) (101-14-4)	I								
Methylmercuric chloride (115-09-3)							I	+(1)	
Methylmercuric hydroxide (1184-57-2)					+(2)				
Methylmercury (22967-92-6)							I		
3-Methyl-4-nitroquinoline 1-oxide (14073-00-8)	I								
Methylnitrosourea (684-93-5)					I				
4-*O*-Methyl-12-*O*-tetradecanoylphorbol 13-acetate (57716-89-9)	I								
Methyluracil (27942-00-3)							I		
Metronidazole (443-48-1)			−(1)						
Mezerein (34807-41-5)	−(1)								
Miconazole (22916-47-8)		+(1)							

TABLE 1 (continued)

Chemical (CAS Registry No.)	Test system [b]								
	Sacch.	Asper.	Neuro.	Plant	Droso.	Mam-CC	Mam-SC	Mam-FG	Mam-MG
Mitomycin C (50-07-7)	I	I	I		−(2)	−(1)	I		I
Morpholine (110-91-8)							I		
MNNG (70-25-7)		W(1)	I	I	+(1)	+(1)			
1-Naphthol (90-15-3)			−(1)						
1-Naphthylamine (134-32-7)	I								
2-Naphthylamine (91-59-8)	I								
Natamycin (7681-93-8)		+(1)							
Neutral red (553-24-2)			−(1)						
Nicotine (54-11-5)			I						
Nitrilotriacetic acid (139-13-9)			−(1)						
4-Nitrobiphenyl (92-93-3)			I						
2-Nitrofluorene (607-57-8)			I						
o-Nitro-*p*-phenylene-diamine (5307-14-2)			I						
p-Nitro-*o*-phenylene-diamine (99-56-9)			I						
4-Nitroquinoline (3741-15-9)			−(1)						
4-Nitroquinoline 1-oxide (56-57-5)	I	+(1)	−(1)						
Nitrosan (2980-64-5)									I
Nitrosomorpholine (59-89-2)	I						I		
Nitrosopiperidine (100-75-4)					I				
Nitrous oxide (10024-97-2)				I					
Nocodazole (31430-18-9)	+(1)								I
Norethindrone acetate (51-98-9)								+(1)	
Nystatin (1400-61-9)			−(1)						
Oil of nutmeg (8008-45-5)									I
Oleic acid (112-80-1)	I								

TABLE 1 (continued)

Chemical (CAS Registry No.)	Test system [b]								
	Sacch.	Asper.	Neuro.	Plant	Droso.	Mam-CC	Mam-SC	Mam-FG	Mam-MG
Pheniramine maleate (132-20-7)									I
Phenobarbital (50-06-6)									I
Phenol (108-95-2)									I
1-Phenyl-3,3-dimethyltriazene (7227-91-0)					I				
m-Phenylenediamine (108-45-2)			I						
o-Phenylenediamine (95-54-5)			I						
p-Phenylenediamine (106-50-3)			I						
Phenylmercuric acetate (62-38-4)							I		
Phenylmercuric nitrate (55-68-5)			−(1)						
N-Phenyl-*p*-phenylenediamine HCl (2198-59-6)			I						
Phenytoin (57-41-0)						I	I		
Phorbol (17673-25-5)	I								
Phorbol 13-acetate (32752-29-7)	I								
Phorbol 12,13-didecanoate (24928-17-4)	I								
4α-Phorbol 12,13-didecanoate (27536-56-7)	I								
Phosphonoacetyl-L-aspartic acid (51321-79-0)							I		
Picloram (1918-02-1)		−(1)							
Potassium dichromate (7778-50-9)							I		
Potassium fluoride (7789-23-3)			I						
Primidone (125-33-7)						I			
Procarbazine (366-70-1)					I				I
Proflavin (92-62-6)					+(1)				
Progesterone (57-83-0)				I			I		
β-Propiolactone (57-57-8)	I				I		I		
Propranolol (525-66-6)	I								

TABLE 1 (continued)

Chemical (CAS Registry No.)	Test system [b]								
	Sacch.	Asper.	Neuro.	Plant	Droso.	Mam-CC	Mam-SC	Mam-FG	Mam-MG
Pyrene (129-00-0)	I								
1-(3-Pyridyl-*N*-oxide)-3,3-dimethyltriazene (21600-42-0)					I				
Pyrimethamine (58-14-0)							+(1)	+(1)	
Quercitin (117-39-5)			−(1)						
Quinacrine mustard (4213-45-0)					I				
Resorcinol (108-46-3)			−(1)						
Rhodamine B (81-88-9)						I			
Saccharin (81-07-2)	I		−(1)						
Saccharin sodium (128-44-9)							I		
Safrole (94-59-7)	I								
Sarcosine (107-97-1)			−(1)						
Satratoxin H (53126-64-0)					+(1)				
Shikimic acid (138-59-0)			I						
Simazine (122-34-9)			−(1)		−(1)				
Sodium azide (26628-22-8)				+(1)					
Sodium chloride (7647-14-5)	I								
Sodium cyclamate (139-05-9)					+(1)				
Sodium deoxycholate (302-95-4)		+(1)							
Sodium fluoride (7681-49-4)			I		−(1)				
Sodium nitrate (7631-99-4)							I		
Sodium nitrite (7632-00-0)							I		
Sodium selenite (10102-18-8)									I
Sorbitol (50-70-4)					I				
Stallimycin (636-47-5)						I			
Stannous fluoride (7783-47-3)			I						

TABLE 1 (continued)

Chemical (CAS Registry No.)	Test system [b]								
	Sacch.	Asper.	Neuro.	Plant	Droso.	Mam-CC	Mam-SC	Mam-FG	Mam-MG
Stearic acid (57-11-4)	I								
Styrene (100-42-5)							I		
Sulfacetamide (144-80-9)	I		+(1)						
Sulfanilamide (63-74-1)		I	I						
Sulfur (7704-34-9)									I
Taurodeoxycholic acid (516-50-7)	I								
Terbacil (5902-51-2)			−(1)						
Testosterone (58-22-0)						−(1)			
2,3,7,8-Tetrachlorodibenzo-*p*-dioxin (1746-01-6)							I		
Tetracycline (60-54-8)				+(1)					
12-*O*-Tetradecanoylphorbol (20839-06-9)	I								
12-*O*-Tetradecanoylphorbol 13-acetate (16561-29-8)	+(1)					+(1)			
2,3,5,6-Tetraethylenoimino-1,4-benzoquinone (4239-06-9)					I				
Δ^9-Tetrahydrocannibinol (1972-08-3)							I		I
3,3′,5,5′-Tetramethylbenzidine (54827-17-7)	I								
Thalidomide (50-35-1)					−(1)				
Thiabendazole (148-79-8)		+(1)							I
Thiocid (5964-24-9)	I								
Thiophamine (27079-29-4)		+(1)							
Thio-TEPA (52-24-4)									I
Thiram (137-26-8)		+(1)							
Tolnaftate (2398-96-1)			I						
o-Toluidine (95-53-4)	I		−(1)						
Triaziquone (68-76-8)					−(1)			+(3)	−(1)

TABLE 1 (continued)

Chemical (CAS Registry No.)	Test system [b]								
	Sacch.	Asper.	Neuro.	Plant	Droso.	Mam-CC	Mam-SC	Mam-FG	Mam-MG
Trichlorfon (52-68-6)									I
1,1,1-Trichloroethane (71-55-6)	I								
2,4,5-Trichlorophenol (95-95-4)				W(1)					
2,4,5-Trichloro-phenoxyacetic acid (93-76-5)				W(1)				I	
1-(2,4,6-Trichloro-phenyl)-3,3-dimethyl-triazene (50355-74-3)					I				
4,5,6-Trichloro-2-trifluoromethyl-benzimidazole (2338-27-4)		−(1)							
Triethylenemelamine (51-18-3)					I				
Trifluralin (1582-09-8)		W(1)	+(1)						
Triflupromazine (117-89-5)	I								
Trimethoprim (738-70-5)			+(1)						
Trophosphamide (22089-22-1)					I				
Urethane (51-79-6)	I		−(1)		−(1)				
Vinblastine (865-21-4)					+(1)	+(2)			
Vinblastine sulfate (143-67-9)			−(1)						
Vincristine (57-22-7)									I
Vinyl chloride (75-01-4)					I				
Zinc chloride (7646-85-7)			I						
Zinc oxide (1314-13-2)							I		

[a] This table is a summation of the current data base on the chemical induction of aneuploidy. Experimental details and references are given in the individual working group reports. It should be emphasized that many of the evaluations in this table are each derived from a single paper and should be verified.

[b] Sacch., *Saccharomyces cerevisiae*; Asper., *Aspergillus nidulans*; Neuro., *Neurospora crassa*; Plant, plant systems; Droso., *Drosophila melanogaster* chromosome gain; Mam-CC, mammalian cell culture; Mam-SC, mammalian somatic cells in vivo; Mam-FG, mammalian female germ cells; Mam-MG, mammalian male germ cells. Positive evaluation for mammalian systems required evidence for hyperploidy.

[c] +, positive; W, weak positive; −, negative; I, inconclusive (reasons for inconclusive evaluations are provided in individual working group reports); numbers in parentheses represent the number of acceptable papers reporting this result.

[d] Positive only in the presence of S9 mix.

represented more than once. Some are known DNA-damaging agents and are detectable in assays for mutagenicity and chromosome breakage as well as those for aneuploidy; these may produce changes in chromosome number as a secondary effect of structural chromosome damage. Alkylating agents (e.g., ethyl methanesulfonate, *N*-methyl-*N'*-nitro-*N*-nitrosoguanidine (MNNG), 1,2-dicloroethane, cyclophosphamide) react with nucleophilic sites in protein as well as in DNA, and may produce aneuploidy directly by alkylation of spindle apparatus proteins or kinetochore DNA. Guanosine triphosphate (GTP) is required for tubulin polymerization (Olmsted and Borisy, 1975); preliminary evidence suggests that alkylation of GTP precursors may affect spindle function (Bonatti and Abbondandolo, 1985).

Despite the inclusion of some DNA-damaging agents, the list of compounds that have been found to induce aneuploidy is not a list of standard mutagens. Both classic and less familiar spindle poisons are included; colchicine/colcemid, vinblastine/vincristine, methyl 2-benzimidazole carbamate, nocodazole, griseofulvin and trifluralin (reviewed by Liang and Brinkley, 1985). Diethylstilbestrol affects centrosomes and kinetochores as well as microtubules (Brinkley et al., 1985). Chloral hydrate blocks spindle elongation (Ris, 1949), and diazepam inhibits centriole separation (Andersson et al., 1981). Amphotericin B and deoxycholate are membrane-damaging agents (Bellincampi et al., 1980; Assinder and Upshall, 1982). All of these compounds show at least one positive evaluation in Table 1, providing some evidence for the proposition that no single target structure or mechanism can account for aneuploidy induction.

The significance attributed to aneuploidy as a factor in the induction and progression of neoplasms varies from author to author (Barrett et al., 1985; Evans, 1985). Some animal carcinogens (diethylstilbestrol, methyl 2-benzimidazole carbamate, benomyl) seem to produce aneuploidy as their principal genotoxic effect (Table 1); evidence for the induction of gene mutations or chromosomal aberrations by these compounds is largely negative or equivocal (Dellarco et al., 1985). Standard test strategies, without aneuploidy assays, would miss the genotoxic and carcinogenic potential of these compounds. In the International Study for the Evaluation of Potential Carcinogens (Ashby et al., 1985), aneuploidy assays were able to detect some animal carcinogens that were inactive in other tests for genotoxicity in vitro.

(4) Test performance

The current data base is inadequate for comparing the performance of different assay methods. Reproducibility of results among laboratories using the same assay cannot be determined because results from only one laboratory are available for most compounds. A few compounds can be compared tentatively across different assay systems. Eight agents (chloral hydrate, colcemid, colchicine, griseofulvin, methotrexate, MNNG, 12-*O*-tetradecanoylphorbol-13-acetate and vinblastine) gave positive results both in a mammalian assay and in at least one non-mammalian system. For benomyl and cyclophosphamide, positive results in mammalian assays were concordant with some non-mammalian results and discordant with others. In 2 other cases, benzo[*a*]pyrene and diethylstilbestrol, negative results were obtained in one non-mammalian assay and positive results in one mammalian assay. Halothane was negative in a mammalian assay, but positive in a non-mammalian assay. Three of the instances of discordance (diethylstilbestrol, benomyl and cyclophosphamide) involved negative tests in Neurospora. Because there is considerable evidence for aneuploidy induction by each of these compounds, the incidence of false negative results in the Neurospora assay needs further investigation. One compound gave discordant results in two mammalian assays; triaziquone was positive in female germ cells but a qualified negative in male germ cells. (It was also negative in Drosophila.) Overall, however, these results in Table 1 permit some optimism about the ability of non-mammalian systems to predict aneuploidy in mammals.

Comparisons of mitotic with meiotic aneuploidy induction should be made initially within the same species to avoid possible interspecies variation. Saccharomyces, the mammal, and some plants are currently the only organisms in which this is possible. Of the compounds in Table 1, only one has evaluated results for both mitotic and meiotic cells of the same species. The antimicrobial drug pyri-

methamine induced hyperploidy in rat bone marrow cells (Barilyak et al., 1978) and hyperploidy in rat oocytes (Chebotar, 1980). Mitotic and meiotic test results are illustrated graphically in Waters et al. (1986).

(5) Future directions

Several points have been reiterated throughout this series of reports; (i) the relevance of aneuploidy to human health; (ii) the need for development, standardization and validation of assay procedures; and (iii) the need for a broader and more reliable data base.

New assay methods with improved reliability, accuracy and speed of analysis are needed, particularly for mammalian systems. Standardization of existing and newly developed procedures is a prerequisite for improvement of the data base because data must be comparable among laboratories. This encompasses more than experimental protocols and methods of data collection; criteria for positive and negative tests, an acceptable range of background frequencies, appropriate solvent and positive controls and appropriate methods for statistical analysis are needed for each system. The work of development and validation could be facilitated by establishment of a reference set of chemical agents for assessing test performance. Ideally, this set would include compounds known to affect specific parts of the mitotic/meiotic apparatus: microtubules and/or their associated proteins, centrosomes, kinetochores, membranes with associated energy-transducing systems, and, in meiotic cells, the synaptonemal complex. Such agents are potential positive control compounds as well as assay-building tools. Clearly, our knowledge of mechanisms and chemical effects is insufficient to produce a complete list immediately, but it is possible to begin. Colcemid, vinblastine, griseofulvin, and *p*-fluorophenylalanine are classic spindle inhibitors; taxol and chloral hydrate inhibit spindle function without destabilizing microtubules. Ethidium bromide and diazepam affect centrioles; amphotericin B is a membrane-specific agent. These and other possible test compounds and their targets are discussed in detail by Liang and Brinkley (1985). A chemical repository to provide samples of selected compounds to researchers for test development and validation is in the planning stage.

The development of improved assays and the generation of an extensive data base will contribute to better understanding of the underlying mechanisms of chromosome segregation and aneuploidy induction; this, in turn, will improve our ability to predict and estimate the human risk associated with environmental, occupational or therapeutic exposure to chemicals. In this endeavor, as in many others, basic science and applied science are close confederates.

References

Allen, J.W., J.C. Liang, A.V. Carrano and R.J. Preston (1986) Review of literature on chemical-induced aneuploidy in mammalian male germ cells, Mutation Res., 167, 123–137.

Andersson, L.C., V.-P., Lehto, S. Stenman, R.A. Badley and I. Virtanen (1981) Diazepam induces mitotic arrest at prometaphase by inhibiting centriolar separation, Nature (London), 291, 247–248.

Ashby, F., F.J. de Serres, M. Draper, M. Ishidate Jr., B.H. Margolin, B. Matter and M. Shelby (1985) Overview and conclusions of the OPCS collaborative study on in vitro assays, in: J. Ashby and F.J. de Serres (Eds.), Progress in Mutation Research, Vol. 5, Elsevier/North-Holland, Amsterdam, pp. 117–174.

Assinder, S.J., and A. Upshall (1982) Mitotic aneuploidy induced by sodium deoxycholate in *Aspergillus nidulans*, Mutation Res., 93, 101–108.

Athwal, R.S., and S.S. Sandhu (1985) Measurement of chemically induced mitotic nondisjunction in mammalian cells using a monochromosomal human/mouse somatic cell hybrid, Abstract presented at the Symposium on Aneuploidy: Etiology and Mechanisms, Carnegie Institution, Washington, DC, March 25–29, 1985.

Barilyak, I.R., L.V. Neumerzhitskaya and A. Turkevitch (1978) Antimutagenic and antiteratogenic properties of dimexide (DMSO), Tsitol. Genet. 12(1), 50–56.

Barrett, J.C., M. Oshimura, N. Tanaka and T. Tsutsu (1985) Role of aneuploidy in early and late stages of neoplastic progression of Syrian hamster embryo cells in culture, in: V.L. Dellarco, P.E. Voytek and A. Hollaender (Eds.), Aneuploidy: Etiology and Mechanisms, Plenum, New York, in press.

Bellincampi, D., G. Gualandi, E. LaMonica, C. Poley and G.P. Morpurgo (1980) Membrane-damaging agents cause mitotic non-disjunction in *Aspergillus nidulans*, Mutation Res., 79, 169–172.

Bonatti, S., and A. Abbondandolo (1985) Spindle disfunction caused by treatment of mammalian cells with O^6-ethylguanine, Abstract presented at the Symposium on Aneuploidy: Etiology and Mechanisms, Carnegie Institution, Washington DC, March 25–29, 1985.

Bond, D.J., and Chandley, A.C. (1983) Aneuploidy, Oxford Monographs on Medical Genetics, No. 11, Oxford University Press, London, 198 pp.

Brandriff, B., L. Gordon, L. Ashworth, G. Watchmaker, A. Carrano and A. Wyrobek (1984) Chromosomal abnormalities in human sperm: Comparisons among four healthy men, Hum. Genet., 66, 193.

Brinkley, B.R., A. Tousson and M.M. Valdivia (1985) The kinetochore of mammalian chromosomes: Structure and function in normal mitosis and aneuploidy, in: V.L. Dellarco, P.E. Voytek and A. Hollaender (Eds.), Aneuploidy: Etiology and Mechanisms, Plenum, New York, in press.

Cattanach, B.M., D. Papworth and M. Kirk (1984) Genetic tests for autosomal non-disjunction and chromosome loss in mice, Mutation Res., 126, 189–204.

Chebotar, N.A. (1980) Cytogenetic and morphogenetic changes in oogenesis and embryogenesis of albino rats induced by chloridin and 2,4,5-trichlorophenoxyacetic acid in the preovulation phase of meiosis. Soviet Genet., 16, 781–787.

Cimino, M.C., R.R. Tice and J.C. Liang (1986) Aneuploidy in mammalian somatic cells in vivo, Mutation Res., 167, 107–122.

Dellarco, V.L., K.H. Mavournin and R.R. Tice (1985) Aneuploidy and health risk assessment: Current status and future directions, Environ. Mutagen., 7, 405–424.

Dellarco, V.L., K.H. Mavournin and M.D. Waters (1986) An introduction to a series of U.S. Environmental Protection Agency special committee reports on testing approaches for the detection of chemically induced aneuploidy, Mutation Res., 167, 3–7.

Evans, H.J. (1985) Neoplasia and cytogenetic abnormalities, in: V.L. Dellarco, P.E. Voytek and A. Hollaender (Eds.), Aneuploidy: Etiology and Mechanisms, Plenum, New York, in press.

Fulton, A.M., and D.J. Bond (1984) Dimethylsulfoxide induces aneuploidy in a fungal test system, Mol. Gen. Genet., 197, 347–349.

Galloway, S.M., and J.L. Ivett (1986) Chemically induced aneuploidy in mammalian cells in culture, Mutation Res., 167, 89–105.

Griffiths, A.J.F., H.E. Brockmann, D.M. DeMarini and F.J. de Serres (1986) The efficacy of Neurospora in detecting agents that cause aneuploidy, Mutation Res., 167, 35–45.

Ivett, J.L., and B.M. Brown (1985) An assay to specifically assess induced aneuploidy and/or polyploidy in Chinese hamster ovary (CHO) cells, Abstract presented at the Symposium on Aneuploidy: Etiology and Mechanisms, Carnegie Institution, Washington, DC, March 25–29, 1985.

Käfer, E. (1984) Disruptive effects of ethyl alcohol on mitotic chromosome segregation in diploid and haploid strains of *Aspergillus nidulans*, Mutation Res., 135, 53–75.

Käfer, E., B.R. Scott and A. Kappas (1986) Systems and results of tests for chemical induction of mitotic malsegregation and aneuploidy in *Aspergillus nidulans*, Mutation Res., 167, 9–34.

Liang, J.C., and B.R. Brinkley (1985) Chemical probes and possible targets for the induction of aneuploidy, in: V.L. Dellarco, P.E. Voytek and A. Hollaender (Eds.), Aneuploidy: Etiology and Mechanisms, Plenum, New York, in press.

Liang, J.C., and T.C. Hsu (1985) Correlation between chromosome univalents and aneuploidy in mutagen-treated mouse spermatocytes, Abstract presented at the Symposium on Aneuploidy: Etiology and Mechanisms, Carnegie Institution, Washington, DC, March 25–29, 1985.

Liang, J.C., and K.L. Satya-Prakash (1985) Induction of aneuploidy by mitotic arrestants in mouse bone marrow, Mutation Res., 155, 61–70.

Mailhes, J.B., R.J. Preston and K.S. Lavappa (1986) Mammalian in vivo assays for aneuploidy in female germ cells, Mutation Res., 167, 139–148.

Martin, R.H. (1984) A detailed method for obtaining preparations of human sperm chromosomes, Cytogenet. Cell Genet., 35(4), 253–256.

Mason, J.M., and M.A. Resnick (1985) Mechanisms and detection of chromosome malsegregation using Drosophila and the yeast *Saccharomyces cerevisiae*, in: V.L. Dellarco, P.E. Voytek and A. Hollaender (Eds.), Aneuploidy: Etiology and Mechanisms, Plenum, New York, in press.

Olmsted, J.B., and G.G. Borisy (1975) Ionic and nucleotide requirements for microtubule polymerization in vitro, Biochemistry, 14, 2996–3005.

Redei, G.P., E.R. Sears, C. Hall and S.S. Sandhu (1985) A novel method for the detection of somatic aneuploidy in intact leaves of hexaploid wheat, Abstract presented at the Symposium of Aneuploidy: Etiology and Mechanisms, Carnegie Institution, Washington, DC, March 25–29, 1985.

Resnick, M.A, V.W. Mayer and F.K. Zimmermann (1986) The detection of chemically induced aneuploidy in *Saccharomyces cerevisiae*: An assessment of mitotic and meiotic systems, Mutation Res., 167, 47–60.

Ris, H. (1949) The anaphase movement of chromosomes in the spermatocytes of the grasshopper, Biol. Bull., 96, 90–106.

Russell, L.B. (1976) Numerical sex-chromosome anomalies in mammals: their spontaneous occurrence and use in mutagenesis studies, in: A. Hollaender (Ed.), Chemical Mutagens, Vol. 4, Plenum. New York, pp. 55–91.

Russell, L.B. (1979) Meiotic nondisjunction in the mouse: Methodology for genetic testing and comparison with other methods, Environ. Health Perspect., 31, 113–128.

Russell, L.B. (1985) Experimental approaches for the detection of chromosomal malsegregation occurring in the germline of mammals, in: V.L. Dellarco, P.E. Voytek and A. Hollaender (Eds.), Aneuploidy: Etiology and Mechanisms, Plenum, New York, in press.

Sandhu, S.S., B.K. Vig and M.J. Constantin (1986) Detection of chemically induced aneuploidy with plant test systems, Mutation Res., 167, 61–69.

Sugawara, S., and K. Mikamo (1983) Absence of correlation between univalent formation and meiotic nondisjunction in aged female Chinese hamsters, Cytogenet. Cell Genet., 35, 34–40.

Tanzarella, C., F. Degrassi and A. Guarino (1985) The in vitro micronucleus assay for the detection of chromosome aneuploidy, Abstract presented at the Symposium on Aneuploidy: Etiology and Mechanisms, Carnegie Institution, Washington, DC, March 25–29, 1985.

Tsutsui, T., H. Maizumi and J.C. Barrett (1984) Colcemid-induced neoplastic transformation and aneuploidy in Syrian hamster embryo cells. Carcinogenesis, 5, 89–93.

Vig, B.K. (1975) Soybean (*Glycine max*) a new test system for study of genetic parameters as affected by environmental mutagens, Mutation Res., 31, 49–56.

Waters, M.D., H.F. Stack, K.H. Mavournin and V.L. Dellarco (1986) Genetic activity profiles of chemicals selected from the aneuploidy data base, Mutation Res., 167, 171–189.

Wyrobek, A., G. Watchmaker, L. Gordon, A.V. Carrano, L. Ashworth and B. Brandriff (1984) Aneuploidy and quinacrine-positive spots in human sperm, Am. J. Hum. Genet., 36, 1183.

Zimmering, S., J.M. Mason and C. Osgood (1986) Current status of aneuploidy testing in Drosophila, Mutation Res., 167, 71–87.

Zimmermann, F.K., V.W. Mayer and I. Scheel (1984) Induction of aneuploidy by oncodazole (nocodazole), an antitubulin agent, and acetone, Mutation Res., 141, 15–18.

Zimmermann, F.K., V.W. Mayer, I. Scheel and M.A. Resnick (1985) Acetone, methyl ethyl ketone, ethyl acetate, acetonitrile and other polar aprotic solvents are strong inducers of aneuploidy, Mutation Res., 149, 339–351.

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